(i) transforming cells of a tomato plant with a plant cell expression vector having an *E. coli* origin or replication, an enhancer, a selectable marker-encoding nucleotide sequence operably linked to a promoter effective to express the selectable marker encoding sequence, a transcription termination element for said selectable marker-encoding nucleotide sequence and a T-DNA sequence in a manner effective to express said selectable marker-encoding nucleotide sequence;

(ii) selecting plant cells which have been transformed by their ability to grow in the presence of an amount of selective agent that is toxic to non-transformed plant cells;

(iii) regenerating transformed plant cells to yield mature plants;

(iv) selecting plants having a desired trait; and

(v) identifying, isolating and characterizing genes the transcription of which was enhanced by said element which functions to enhance gene expression.

21. (New) The method of claim 20 wherein said transforming cells is by introduction of *Agrobacterium tumifaciens* into hypocotyls derived from said tomato plant in the absence of feeder cells.

22. (New) The method of claim 20 wherein said transforming cells is by introduction of *Agrobacterium tumifaciens* into shoot tip tissue derived from said tomato plant in the absence of feeder cells.

23. (New) The method of claim 20 wherein said tomato plant is a dwarf tomato plant.

Please cancel claims 12, 14, 15 and 18 without prejudice.

Remarks

Upon entry of this amendment, claims 1-11 and 16-23 will be pending.

Claims 1, 3-7, 11 and 19 have been amended to more particularly point out and distinctly claim the subject matter, which Applicants regard as the invention.

Specifically, Claim 1 has been amended to recite the features that the method encompassed by the claim comprises transforming cells of fleshy fruit-bearing plant. Support for this feature is found *inter alia* on page 2 lines 21-22 and page 14 line 41 – page 15 line 5. Claim 11, which depends upon claim 1, and claim 19, which encompasses the method of claim 1, have been similarly amended to recite the feature of a fleshy fruit-bearing plant in order to maintain proper antecedence. Claim 1 has further been amended to remove the feature that " transforming cells is by introduction of *Agrobacterium tumifaciens* into hypocotyls or shoot tip tissue derived from said plant in the absence of feeder cells."

Claim 3 has been amended to substitute the term "enhancer" for the phrase "element which functions to enhance gene expression," in order to maintain proper antecedence with this feature in claim 1, upon which Claim 3 depends. In the previously filed amendment to claim 1 (filed August 6, 2001), the phrase "element which functions to enhance gene expression" was replaced with "enhancer."

Claim 3 has further been amended to specify that the enhancer may be a Figwort Mosaic Virus (FMV) enhancer or the mirabilis mosaic virus (MMV) enhancer. Support for the "Figwort Mosaic Virus (FMV) enhancer" feature is found on page 12 lines 40-41 and in claim 5, as originally filed. Support for the "mirabilis mosaic virus (MMV) enhancer" feature is found on page 13 lines 10-13. Additionally, the feature that the MMV promoter sequence is the sequence of SEQ ID NO:8 has been removed.

Claim 4 has been amended to narrow the scope of the claim and now specifies that the enhancer has the sequence presented as SEQ ID NO:1.

Claim 5 has been amended to narrow the scope of the claim and now specifies that the enhancer has the sequence presented as SEQ ID NO:5 or SEQ ID NO:6. Claim 5 has been further amended to specify that the sequence of SEQ ID NO:6 is Figwort Mosaic Virus (FMV) enhancer.

Claim 6 has been amended to narrow the scope of the claim and now specifies that the enhancer has the sequence presented as SEQ ID NO:7.

Claim 7 has been amended to specify that the enhancer is either the MMV promoter presented as SEQ ID NO:8 or the MMV enhancer presented as nucleotides 1-260 of SEQ ID NO:8. Support for the feature that the enhancer may be the MMV enhancer presented as nucleotides 1-260 of SEQ ID NO:8 is found on page 13 lines 10-13.

Claim 2 has been amended to correct a typographical error. Specifically, two steps had been labeled "vii;" the second of these has now been correctly labeled "viii."

Independent claim 20 and dependent claims 21-23 have been added to more particularly point out and distinctly claim the subject matter that applicants regard as the invention. Specifically, claim 20 recites a method of method for identifying genes associated with a desired trait in a tomato plant. Support for claim 20 is found, *inter alia*,

on page 15 lines 3-6 and in claim 1, as originally filed. Further support is found in the Examples, page 21 line 12 – page 24 line 38.

Claim 21 specifies that the "transforming cells" step in the method of claim 20 is by introduction of *Agrobacterium tumifaciens* into hypocotyls derived from tomato plant in the absence of feeder cells. Claim 22 specifies that the "transforming cells" step in the method is by introduction of *Agrobacterium tumifaciens* into shoot tip tissue derived from tomato plant in the absence of feeder cells. Support for these features is found, *inter alia*, on page 16 lines 5-10 and in Example 1, page 21 line 13 – page 22 line 19.

Claim 23 specifies that the tomato plant is a dwarf tomato plant; support for this feature is found, *inter alia*, on page 15 lines 11-24.

No new matter is introduced by the amendments to the claims. A marked-up copy of the amended claims is provided in Appendix A. A clean version of the entire set of pending claims is provided in Appendix B. .

REPLY

Claim Rejections – 35 USC § 112

The examiner has rejected claim 18 under 35 USC § 112, second paragraph, and claims 14 and 15 under 35 USC § 112, second paragraph. These three claims have been cancelled.

Claim Rejections – 35 USC § 103

The Examiner has rejected claims 1-12 and 14-19, allegedly as being unpatentable over Hayashi *et al.*, (Science 1992, 258:1350-1353) in view of Schell *et al.* (The Plant Journal 1999, 17:461-466). In response to applicants' previous argument that the taught method has not been practical in "fruit-bearing plants," the examiner has asserted that fruit-bearing plants encompass all dicotyledonous plant species, such as tobacco, which produces a capsule fruit. Applicants have amended claim 1, and all claims that depend therefrom, to specifically recite the feature of a "fleshy fruit-bearing plant." It is believed that this amendment will overcome the examiner's rejection based on the premise that the claim encompasses all dicotyledonous plant species that produce capsule fruit. The meaning of "fleshy fruit" is well known in the art. For instance, the *Dictionary of Science and Technology* (Morris C (ed.), Academic Press, Harcourt Brace Jovanovich Publishers,

San Diego, 1992) defines "fleshy fruit" as: "any fruit having juicy, pulpy cellular tissue, such as peaches or melons." (The full text of this definition, accompanied by publication information for the *Dictionary*, is included herewith as an Exhibit.) Moreover, applicants have specified that the methods of the invention are directed to fleshy fruit bearing plants. On page 14 line 41 – page 15 line 2, is stated:

"Plants for use in carrying out the trait-associated gene identification methods of the invention must have the following properties: ... (3) the ability to produce a fleshy fruit..."

It is noted that the examiner did not address the basis for including in the rejection dependent claims 11 and 12, which are directed to gene identification in dwarf plants and dwarf tomato plants (although applicants currently request cancellation of claim 12, new claims 20 and 23 are directed to similar methods in tomato and dwarf tomato plants). The Examiner has argued that the Schell reference teaches that other researchers have successfully applied the technique in identifying other genes in other plant species, specifically calling attention to page 462, right column. However, the references provided in Schell only describe this method in tobacco and *Arabidopsis*. If the Examiner is implying that it would be *prima facie* obvious to apply activation tagging methodologies to all plant species, applicants respectfully traverse the rejection. On this subject, § M.P.E.P. 2143.03 teaches: "To establish *prima facie* obviousness of a claimed invention, all claim limitations must be taught or suggested by the prior art." The Examiner has cited no references that apply to dwarf plants, nor dwarf tomato plants. Thus, it is unclear to applicants what the basis is for rejecting claims 11 and 12. Applicants further point out that the Examiner has provided no evidence that suggests the validity of the method in fleshy fruit-bearing species, which applicants maintain is a novel and non-obvious feature of the invention.

The Examiner has further argued that the use of hypocotyls and shoot tip tissues were well known in the art at the time of applicants' invention (although the instant amendment requests removal of this feature from claim 1, new claims 21 and 22 recite essentially the same feature). However, the claim limitation that this argument addresses does not merely recite transformation using these particular tissues, but, more importantly, it recites a method of transforming these tissues in the absence of feeder cells. Again, applicants submit that the Examiner has failed to make a case of *prima facie* obviousness since he has not addressed every claim limitation. Applicants moreover maintain that the claimed methods of transforming tomato cells by co-cultivation in the absence of feeder cells was novel and non-obvious over the prior art

(represented by the 1996 Frary and Earle reference) at the time of applicants' invention, and that these method provided simpler and more efficient transformation techniques.

The Examiner has rejected claim 19, asserting that the transgenic plant of this claim remains is obvious in view of Hayashi (1992). Applicants have amended claim 19 to specifically recite a "transgenic fleshy fruit-bearing plant." It is believed that the instant amendment to claim 19 obviates the basis for this rejection, since neither Hayashi nor Schell disclose methods pertaining to fleshy fruit bearing plants. Applicants assert that fleshy fruit-bearing plants displaying the morphological characteristics recited in claim 19 are novel and non-obvious over the prior art.

CONCLUSION

In view of the claim amendments and for the above reasons, it is believed that all of the rejections are overcome, and that the claims are in condition for allowance.

Respectfully submitted,

Dated: April 22, 2002

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APPENDIX A

1. (Twice amended) A method for identifying genes associated with a desired trait in a fleshy fruit-bearing plant comprising:

(i) transforming cells of a fleshy fruit-bearing plant with a plant cell expression vector having an *E. coli* origin or replication, an enhancer, a selectable marker-encoding nucleotide sequence operably linked to a promoter effective to express the selectable marker encoding sequence, a transcription termination element for said selectable marker-encoding nucleotide sequence, and a T-DNA sequence [, wherein said transforming cells is by introduction of *Agrobacterium tumifaciens* into hypocotyls or shoot tip tissue derived from said plant in the absence of feeder cells,] in a manner effective to express said selectable marker-encoding nucleotide sequence;

(ii) selecting plant cells which have been transformed by their ability to grow in the presence of an amount of selective agent that is toxic to non-transformed plant cells;

(iii) regenerating transformed plant cells to yield mature plants;

(iv) selecting plants having a desired trait; and

(v) identifying, isolating and characterizing genes the transcription of which was enhanced by said element which functions to enhance gene expression.

2. (Twice Amended) The method of claim 1, further comprising the steps of

(vi) preparing a separate heterologous gene construct for each isolated gene;

(vii) transforming plants with said separate heterologous gene construct wherein expression of the isolated gene is enhanced in said plants;

(viii [vii]) selecting plants having the desired trait.

3. (Amended) The method of claim 1, wherein the enhancer [element which functions to enhance gene expression] is selected from the group consisting of a CaMV35S enhancer element, a Figwort Mosaic Virus (FMV) promoter[sequence], a Figwort Mosaic Virus (FMV) enhancer, a peanut chlorotic streak caulimovirus full-length transcript (PClSVFLt) sequence, [and] a mirabilis mosaic virus (MMV) [,] promoter and a mirabilis mosaic virus (MMV) enhancer [sequence (SEQ ID NO:8)].

4. (Twice amended) The method of claim 3, wherein said enhancer is the CaMV35S enhancer element, and wherein the CaMV35S enhancer element is a 4X tandem duplicated CaMV35S enhancer element having the sequence presented as SEQ ID NO:1.

5. (Twice amended) The method of claim 3, wherein said enhancer is the Figwort Mosaic Virus (FMV) promoter [sequence is the promoter] having the sequence presented as SEQ ID NO:5 or the Figwort Mosaic Virus (FMV) enhancer having the sequence presented as SEQ ID NO:6.

6. (Twice amended) The method of claim 3, wherein said enhancer is the peanut chlorotic streak caulimovirus full-length transcript (PClSVFLt) sequence, and wherein the PClSVFLt sequence is the enhancer having the sequence presented as SEQ ID NO:7.

7. (Twice amended) The method of claim 3, wherein said enhancer is the mirabilis mosaic virus (MMV) promoter [sequence is the promoter] having the sequence presented as SEQ ID NO:8 or the mirabilis mosaic virus (MMV) enhancer having the sequence presented as nucleotides 1-260 of SEQ ID NO:8.

11. (Amended) The method of claim 1, wherein said fleshy fruit-bearing plant is a dwarf plant.

19. (Amended) A transgenic fleshy fruit-bearing plant comprising enhanced expression of a gene identified by the method of claim 1, wherein the gene is associated with a morphological characteristic selected from the group consisting of leaflet size, leaf size, leaf color, leaf shape, leaflet number, leaf number, internode length, plant height, floral organ characteristics and fruit characteristics.

APPENDIX B

1. A method for identifying genes associated with a desired trait in a fleshy fruit-bearing plant comprising:

(i) transforming cells of a fleshy fruit-bearing plant with a plant cell expression vector having an *E. coli* origin or replication, an enhancer, a selectable marker-encoding nucleotide sequence operably linked to a promoter effective to express the selectable marker encoding sequence, a transcription termination element for said selectable marker-encoding nucleotide sequence, and a T-DNA sequence, wherein said transforming cells is by introduction of *Agrobacterium tumifaciens* into hypocotyls or shoot tip tissue derived from said plant in the absence of feeder cells, in a manner effective to express said selectable marker-encoding nucleotide sequence;

(ii) selecting plant cells which have been transformed by their ability to grow in the presence of an amount of selective agent that is toxic to non-transformed plant cells;

(iii) regenerating transformed plant cells to yield mature plants;

(iv) selecting plants having a desired trait; and

(v) identifying, isolating and characterizing genes the transcription of which was enhanced by said element which functions to enhance gene expression.

2. The method of claim 1, further comprising the steps of

(vi) preparing a separate heterologous gene construct for each isolated gene;

(vii) transforming plants with said separate heterologous gene construct wherein expression of the isolated gene is enhanced in said plants;

(viii) selecting plants having the desired trait.

3. The method of claim 1, wherein the enhancer is selected from the group consisting of a CaMV35S enhancer element, a Figwort Mosaic Virus (FMV) promoter, a Figwort Mosaic Virus (FMV) enhancer, a peanut chlorotic streak caulimovirus full-length transcript (PClSVFLt) sequence, a mirabilis mosaic virus (MMV) promoter and a mirabilis mosaic virus (MMV) enhancer.

4. The method of claim 3, wherein said enhancer is the CaMV35S enhancer element, and wherein the CaMV35S enhancer element is a 4X tandem duplicated CaMV35S enhancer element having the sequence presented as SEQ ID NO:1.

5. The method of claim 3, wherein said enhancer is the Figwort Mosaic Virus (FMV) promoter having the sequence presented as SEQ ID NO:5 or the Figwort Mosaic Virus (FMV) enhancer having the sequence presented as SEQ ID NO:6.

6. The method of claim 3, wherein said enhancer is the peanut chlorotic streak caulimovirus full-length transcript (PClSVFLt) sequence, and wherein the PClSVFLt sequence is the enhancer having the sequence presented as SEQ ID NO:7.

7. The method of claim 3, wherein said enhancer is the mirabilis mosaic virus (MMV) promoter having the sequence presented as SEQ ID NO:8 or the mirabilis mosaic virus (MMV) enhancer having the sequence presented as nucleotides 1-260 of SEQ ID NO:8.

8. The method of claim 1, wherein said selectable marker-encoding nucleotide sequence encodes a polypeptide which confers herbicide-resistance to transformed plant cells expressing said marker.

9. The method of claim 1, wherein said selectable marker-encoding nucleotide sequence encodes an antibiotic resistance gene which confers resistance to an antibiotic selected from the group consisting of kanamycin, G418, bleomycin, hygromycin, chloramphenicol, ampicillin and tetracycline.

10. The method of claim 9, wherein said antibiotic is kanamycin.

11. The method of claim 1, wherein said fleshy fruit-bearing plant is a dwarf plant.

16. The method of claim 1, wherein said desired trait is a biochemical modification of a plant and fruit selected from the group consisting of a change in the level of vitamins minerals, elements, amino acids, carbohydrates, lipids, nitrogenous bases, isoprenoids, phenlypropanoids and alkaloids.

17. The method of claim 1, wherein said desired trait is a fruit-bearing plant specific trait selected from the group consisting of increased resistance to fungal pathogens, increased resistance to bacterial pathogens, increased resistance to viral pathogens, increased resistance to insects, modified flower size, modified flower number, modified flower pigmentation, modified flower shape, modified leaf number, modified leaf pigmentation, modified flower shape, modified seed number, a modified pattern of leaves and flowers, a modified distribution of leaves and flowers, modified stem length between nodes, modified root mass, increased drought tolerance, increased salt tolerance and increased antibiotic tolerance.

19. A transgenic fleshy fruit-bearing plant comprising enhanced expression of a gene identified by the method of claim 1, wherein the gene is associated with a morphological characteristic selected from the group consisting of leaflet size, leaf size, leaf color, leaf shape, leaflet number, leaf number, internode length, plant height, floral organ characteristics and fruit characteristics.

20. A method for identifying genes associated with a desired trait in a tomato plant comprising:

(i) transforming cells of a tomato plant with a plant cell expression vector having an *E. coli* origin or replication, an enhancer, a selectable marker-encoding nucleotide sequence operably linked to a promoter effective to express the selectable marker encoding sequence, a transcription termination element for said selectable marker-encoding nucleotide sequence and a T-DNA sequence in a manner effective to express said selectable marker-encoding nucleotide sequence;

(ii) selecting plant cells which have been transformed by their ability to grow in the presence of an amount of selective agent that is toxic to non-transformed plant cells;

(iii) regenerating transformed plant cells to yield mature plants;

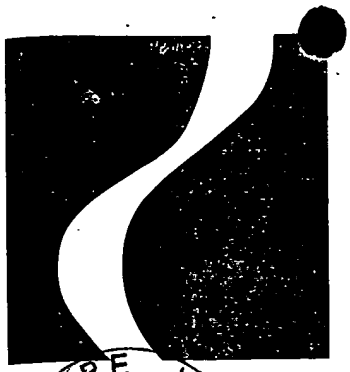
(iv) selecting plants having a desired trait; and

(v) identifying, isolating and characterizing genes the transcription of which was enhanced by said element which functions to enhance gene expression.

21. The method of claim 20 wherein said transforming cells is by introduction of *Agrobacterium tumifaciens* into hypocotyls derived from said tomato plant in the absence of feeder cells.

22. The method of claim 20 wherein said transforming cells is by introduction of *Agrobacterium tumifaciens* into shoot tip tissue derived from said tomato plant in the absence of feeder cells.

23. The method of claim 20 wherein said tomato plant is a dwarf tomato plant.



Academic Press Dictionary of Science and Technology

Edited by
Christopher Morris

Academic Press
Harcourt Brace Jovanovich, Publishers
San Diego New York Boston London Sydney Tokyo Toronto

Exhibit

This book is printed on acid-free paper. ♾

Academic Press, Inc.
1250 Sixth Avenue, San Diego, California 92101-4311

United Kingdom Edition published by
Academic Press Limited
24–28 Oval Road, London NW1 7DX

Library of Congress Cataloging-in-Publication Data

Academic Press dictionary of science and technology / edited by
Christopher Morris
p. cm.
ISBN 0-12-200400-0
1. Science--Dictionaries. 2. Technology--Dictionaries.
I. Morris, Christopher G. II. Academic Press. III. Title:
Dictionary of science and technology.
Q123.A33 1991
503--dc20 90-29032
CIP

PRINTED IN THE UNITED STATES OF AMERICA
92 93 94 95 96 97 DO 9 8 7 6 5 4 3 2 1

fleece *Vertebrate Zoology.* the coat of wool that covers a sheep or similar animal. *Textiles.* any soft fabric resembling a sheep's coat.

fleet *Military Science.* an organization of ships, aircraft, marine forces, and shore-based activities under a single commander who may exercise operational as well as administrative control. *Transportation Engineering.* the total number of vehicles or cars in a system, including active and inactive units. *Mechanical Engineering.* the sideways movement of a rope or cable when being wound on a drum.

fleet angle *Mechanical Engineering.* the maximum angle between a rope or cable and a line drawn perpendicular to the drum on which it winds.

fleet ballistic missile *Ordnance.* a submarine or shipborne strategic ballistic missile; e.g., Polaris or Poseidon. Thus, **fleet ballistic missile submarine.**

fleet broadcast *Telecommunications.* the radio broadcast received by all U.S. Navy ships and merchant vessels, consisting mostly of weather bulletins.

fleeting target *Ordnance.* a moving target that remains within observation or firing distance for such a short duration that it is difficult to take deliberate aim.

Fleming, Sir Alexander 1881–1955, Scottish bacteriologist; discovered lysozyme; shared Nobel Prize for development of penicillin.

Alexander Fleming

Fleming, John Ambrose 1849–1945, English electrical engineer; pioneer in electrical heating and lighting; invented Fleming radio rectifier.

Fleming, Sir Sandford 1827–1915, Canadian civil engineer; coordinated development of railways, telegraph, and standard time zones.

Fleming cracking process *Chemical Engineering.* a thermal cracking method for heavy petroleum fractions in a vertical shell still under pressure.

Fleming tube *Electronics.* a diode that allows negative current to flow from its heated filament to its cold electrode, but prevents it from flowing in the reverse direction.

Flemish bond *Building Engineering.* a masonry bond that consists of alternate headers and stretchers in every course, with each header centering on the stretchers in the courses above and below.

Flemish coil *Naval Architecture.* a rope coiled flat in tight concentric circles so that it resembles a circular mat.

Flemish garden wall bond *Building Engineering.* a masonry bond consisting of stretches and headers in the ratio of one to three or four in each course, with joints broken to give varied patterns.

flesh *Anatomy.* the soft muscular tissue of the body.

Flesh-Demag process *Chemical Engineering.* a procedure for making gas using a cyclic water-gas unit for feeding, and charring coal for gas generation.

flesher *Archaeology.* a long, broad-edged tool of bone, antler, or stone that is used to scrape or rub hides free of fat, sinew, hair, and other unwanted material.

fleshing machine *Mechanical Engineering.* equipment that strips flesh from hides in a tannery.

fleshy fruit *Botany.* any fruit having juicy, pulpy cellular tissue, such as peaches or melons.

Fletcher-Munson curve *Acoustics.* a graph plotting sound intensity, as expressed in decibels, in comparison with frequency, for which each point represents equal loudness over the contour. Also, **Fletcher-Munson contour.**

Fletcher radial burner *Engineering.* a burner whose gas jets are in a radial configuration.

Fleury's algorithm [flůr'ēz] *Mathematics.* a method for constructing an Eulerian path in a given Eulerian graph, as follows: Start at any vertex and traverse the edges in an arbitrary manner. Erase the edges as they are traversed, and if any isolated vertices result, erase them too. At each stage, use an isthmus only if there is no alternative. The result is an Eulerian path.

flex *Mechanics.* **1.** to move from a straight course or position; bend or curve. **2.** the act of bending, or the capacity to bend.

flexed burial *Archaeology.* a method of burial in which the body is interred in a fetal position.

Flexibacter *Bacteriology.* a genus of gliding, rod-shaped bacteria of the family Cytophagaceae, occurring in soil, freshwater, or marine habitats; some species are pathogenic for fish.

Flexibacteriae *Bacteriology.* a class of gliding bacteria that includes the orders Cytophagales and Myxobacterales.

Flexibilia *Paleontology.* a small subclass of crinoids in the subphylum Crinozoa, extant from the Ordovician to the Permian; characterized by loosely joined calycal plates.

flexibility *Mechanics.* **1.** the quality or condition of offering little resistance to being bent; being able to withstand repeated bending. **2.** see ELASTICITY. *Robotics.* the ability of a robot to bend or flex repeatedly through the use of joints and links.

flexible *Mechanics.* having the property of flexibility; able to be repeatedly bent and still maintain its original shape afterward. *Design Engineering.* adaptable to a variety of parts and assembly processes. Thus, **flexible assembly, flexible automated manufacturing, flexible cells and systems, flexible fixturing, flexible flow line.**

flexible circuit *Electronics.* a device in which the electrical elements are mounted onto a pliable plastic sheet, so that it may be fitted between large components.

flexible coupling *Mechanical Engineering.* a coupling containing a resilient member such as a metal spring or rubber disk; used to connect two rigid shafts that cannot be aligned. *Electromagnetism.* a connection between two waveguides that allows a limited range of angular movement between axes.

flexible disk see DISKETTE.

flexible DNC *Robotics.* a process of DNC (distributed numerical control) in which a large number of machine tools and automated materials-handling systems are connected to a host computer.

flexible gun *Ordnance.* a gun mounted so that it can be moved both vertically and horizontally; usually applied to a machine gun mounted in an aircraft turret.

flexible-joint pipe *Engineering.* a cast-iron pipe designed to lie under water and withstand movement through several degrees without leakage.

flexible manufacturing system *Robotics.* **1.** a complex system for manufacturing that can be programmed to change from one task to another. **2.** a series of computerized machining workstations throughout a manufacturing plant that provide for the automatic production of a related group of workpieces.

flexible mold *Engineering.* a coating composed of flexible rubber or other elastomeric materials; usually used for casting plastics.

flexible pavement *Civil Engineering.* a road or runway surface that has little tensile strength and is therefore flexible; usually made of bituminous material.

flexible resistor *Electricity.* an insulated, wire-wound resistor that can be bent, coiled, or knotted; it has the appearance of a flexible lead.

flexible route *Transportation Engineering.* a route that changes according to demand or conditions.

flexible sandstone *Geology.* a variety of itacolumite characterized by thin layers of fine-grained materials.

flexible shaft *Mechanical Engineering.* **1.** a shaft that can transmit rotary motion up to an angle of 90°. **2.** any shaft that is made of flexible material. **3.** a shaft whose bearings are designed to allow for minor misalignment.

flexible waveguide *Electromagnetism.* a waveguide that can be bent without significantly altering its electromagnetic properties.